

## REMARKS

By this amendment, Claims 22-24 and 28-30 are canceled without prejudice. Claims 1, 18, 31 and 32 have been amended, and new Claim 33 added. Claims 1-21, 25-27 and 31-33 are presently pending.

Claims 1, 18, 31 and 32 have been amended to include the temperature of the pretreatment condition. Support for this amendment may be found on page 15, lines 20-21.

Claim 18 has also been amended to include a period at the end of the claim.

New Claim 33 is directed to a method for producing an embryo in wheat or barley.

### Claim rejections under 35 U.S.C. 103(a)

The Examiner has rejected Claims 1-12 and 18-21 and Claims 31-32 under 35 USC 103(a) alleging that the subject matter of the claims would have been obvious to a person of skill in the art based on Kreuger et al., and in view of Genovesi et al. applicants disagree and traverse the rejection using the following remarks and comments:

Kreuger et al., disclose the use of AGPs to induce or stimulate embryo formation of somatic embryos, in a process conducted at room temperature (22°C; see Column 3, line 56). None of the examples in Kreuger disclose embryo formation using microspore cultures (androgenesis), which is a fundamentally different developmental system from that using somatic embryos. Only in passing does the patent suggest that the method is generally applicable for anther and microspore cultures (Column 2, lines 30-32). However, there is no teaching, suggestion, or guidance within Kreuger as to how a microspore culture could be stimulated to induce embryo formation. Kreuger does not teach or suggest the addition of AGP to a microspore culture, or that the pretreatment of microspores from about 3°C to about 6°C is effective in embryo production from microspores.

Genovesi et al., disclose a method for the production of monocotyledonous plants. The protocol consists of pretreating a plant composition comprising microspores under conditions that divert microspores from gametophytic development to that of embryo development (column 4, lines 16-29) at a temperature of 10°C (Column 5, lines 32 and 60; preferred range between 8°C and 14°C; Column 5, line 12). There is no suggestion, teaching or motivation in this document of compositions comprising arabinogalactan proteins may be used to induce embryogenesis in plants. Furthermore, there is no indication as to when AGP may be used within a protocol to produce embryos from microspores.

It is submitted that the obviousness rejections made by the Examiner can only be established by combining or modifying the teachings of the prior art to produce the claimed invention where there is some teaching, suggestion, or motivation to do so found either in the references themselves or in the knowledge generally available to one of ordinary skill in the art (*In re Fine*, 837 F.2d 1071, 5 USPQ2d 1596 (Fed. Cir. 1988); *in re Jones*, 958 F.2d 347, 21 USPQ2d 1941 (Fed. Cir. 1992)). In this regard, a person of skill would not be motivated to combine the teachings of Kreuger et al with Genovesi et al., particularly since both documents relate to different aspects of plant development (somatic embryogenesis v. androgenesis, respectively), and the processes take place under different conditions (22°C v. 10°C, respectively).

The Genovesi patent is directed to “subjecting plant compositions to a combination of stresses” (column 4, lines 18-19). Specifically, “the pretreatment includes incubation of the plant composition comprising microspores at a cold temperature, which is a stress factor” (column 4, lines 25-26). As would be evident to a person of skill in the art, stress factors are known to activate particular genes within plant cells, and the activation of these genes alters the gene expression profile of the plant cell. Applicants submit that one of skill in the art would be unable to predict with reasonable certainty that a composition comprising arabinogalactan protein would induce embryogenesis in microspores following a cold pretreatment as demonstrated in the instant application. Therefore, the subject matter claimed in the instant application is not obvious in view of Kreuger having regard to Genovesi.

Furthermore, applicants submit that neither of the references cited by the Examiner disclose or suggest incubating a micro-spore containing plant segment “under pre-treatment conditions and a temperature from about 3° C to about 6°C to maintain from about 50% to about 100% of microspores at a uninucleate stage of development” as is recited in the claims of the instant application.

Therefore, based on the arguments provided above, applicants assert that Examiner’s rejection of Claims 1-12 and 18-21 and Claims 31-32 under 35 U.S.C. 103(a) based on Genovesi et al., having regard to Kreuger is without merit, and removal of the rejection is requested.

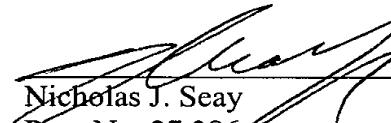
Applicants have cancelled Claim 22 without prejudice. Therefore, the objection to this claim is now moot.

It is respectfully submitted that the above-identified application is now in a condition for allowance and favorable reconsideration and prompt allowance of these claims are

respectfully requested. Should the Examiner believe that anything further is desirable in order to place the application in better condition for allowance, the Examiner is invited to contact the applicants' undersigned attorney at the telephone number listed below.

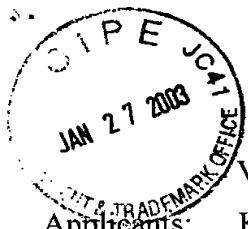
A copy of pages 39-44 of the Hunter reference referred to in the Office Action is enclosed herewith as suggested by the Examiner.

Respectfully submitted,



Nicholas J. Seay  
Reg. No. 27,386  
Attorney for Applicants  
QUARLES & BRADY LLP  
P O Box 2113  
Madison, WI 53701-2113

TEL 608/251-5000  
FAX 608/251-9166



VERSION WITH MARKINGS TO SHOW CHANGES MADE

Applicants: Kenneth J. Kasha  
Ecaterina Simion

Date: January 21, 2003

Serial Number: 09/460,324

Group Art Unit: 1661

Filed: 12/10/99

Examiner: Anne Marie Grunberg

Title: EMBRYOGENESIS AND PLANT  
REGENERATION FROM MICROSPORES

File No.: 411044.90021

In the Claims:

Please amend Claims 1, 18, 31 and 32 and add Claim 33 as follows:

**RECEIVED**  
**JAN 28 2003**  
**TECH CENTER 1600/2900**

1. (Twice amended) A method of producing an embryo comprising the steps of:  
(a) harvesting a microspore-containing plant segment from a donor plant;  
(b) incubating said segment under pre-treatment conditions, and at a temperature from about 3° C to about 6°C, to maintain from about 50% to about 100% of microspores at a uninucleate stage of development;  
(c) isolating microspores from said segment; and  
(d) incubating said isolated microspores in an induction medium comprising arabinogalactan protein, to induce embryogenesis, thereby producing embryos.

18. (Twice Amended) A method of plant regeneration from microspores comprising the steps of:

(a) harvesting a microspore-containing plant segment from a donor plant;  
(b) incubating said segment under pre-treatment conditions, and at a temperature from about 3° C to about 6°C, to maintain from about 50% to about 100% of microspores at a uninucleate stage of development;  
(c) isolating microspores from said segment;  
(d) incubating said isolated microspores in an induction medium comprising an auxin and an arabinogalactan protein, to induce the production of embryos;  
(e) incubating said embryos in a differentiation medium to produce differentiated embryos; and  
(f) regenerating plants from said differentiated embryos.

31. (Amended) A method of producing a composition of microspores comprising:

- (a) harvesting a microspore-containing plant segment from a donor plant;
- (b) incubating said segment under pre-treatment conditions, and at a temperature from about 3° C to about 6°C, to maintain from about 50% to about 100% of microspores at a uninucleate cell cycle;
- (c) isolating microspores from said segment; and
- (d) incubating said isolated microspores in an induction medium comprising an arabinogalactan protein to produce said composition of microspores comprising greater than about 25% viable microspores after a 10 day incubation period.

32. (Amended) A method of producing a composition of microspores comprising:
- (a) harvesting a microspore-containing plant segment from a donor plant;
  - (b) incubating said segment under pre-treatment conditions, and at a temperature from about 3° C to about 6°C, to maintain from about 50% to about 100% of microspores at a uninucleate cell cycle;
  - (c) isolating microspores from said segment; and
  - (d) incubating said isolated microspores in an induction medium comprising an arabinogalactan protein to produce said composition of microspores comprising greater than about 15%.

33. (New) A method of producing an embryo comprising the steps of:
- (a) harvesting a microspore-containing plant segment from a donor wheat or barley plant;
  - (b) incubating said segment under pre-treatment conditions, and at a temperature from about 3° C to about 6°C, to maintain from about 50% to about 100% of microspores at a uninucleate stage of development;
  - (c) isolating microspores from said segment; and
  - (d) incubating said isolated microspores in an induction medium comprising arabinogalactan protein to induce embryogenesis, thereby producing embryos.